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Phytochemical Screening, Proximate and Amino Acid Compositions of Two Edible Indigenous Leafy Vegetables from Ekiti State, Nigeria

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***Abstract*— The leaves of *Albizia zygia* (DC) J. F. Macbr and *Crateva adansonii* DC. were investigated for phytochemical screening, proximate, mineral and amino acid compositions using standard analytical methods. The leaves of the plants were found to contain 29.4% (*Albizia zygia*) and 18.3% (*Crateva adansonii*) crude protein. Their respective crude fat values were 7.63 and 4.11%. while the carbohydrate contents of both leaves were 40.6 (*Albizia zygia*) and 50.6% (*Crateva adansonii*). The phytochemical screening of the methanol extract of *Albizia zygia* and *Crateva adansonii* leaves revealed the presence of alkaloids, tannins, phenols, saponins and flavonoids while steroids and anthraquinones were absent in the two plant leaves. The mineral analysis of the samples showed that calcium had the highest concentration in both samples with *Crateva adansonii* having a higher value of 1210 mg/100g compared to 930mg/100g obtained for *Albizia zygia* leaves. The amino acid composition of the leaves showed that eighteen amino acids were present in both leaves with the highest concentrations recorded for glutamic and aspartic acids. This study has revealed that the two edible indigenous leafy vegetables are a good source of dietary protein, major mineral elements and essential amino acids.**

***Index Terms*- Albizia zygia, amino acids, Crateva adansonii, leaves, phytochemical screening**

I. INTRODUCTION

African leafy vegetables (ALVs) also known as traditional leafy vegetables (TLVs) or indigenous leafy vegetables (ILVs) are defined as leafy green vegetables that have been originally domesticated or cultivated in Africa for the last several centuries [1]. ALVs have long played an important role in the nutrition and diet of sub-Saharan African people. They are generally collected from the wild, with only a few species being cultivated in home gardens or smallholder plots as part of a mixed cropping system [2]. Some researchers have shown that ALVs have micronutrient levels that are comparable to or even higher than those found in most exotic leafy vegetables [3], [4], [5]. Despite the nutritional contribution of ALVs to local diets, and their health maintenance and protective properties [6], most ALVs species are not well known, or they are only used locally and little or no attention has also been given to these vegetables by local, national and international research institutions [7]. *Albizia zygia* and *Crateva adansonii* are examples of indigenous plant trees with edible leaves which are underutilized.

Albizia zygia (DC) J. F. Macbr (*A. zygia*) is a medium-sized gum-producing tree that is widespread in tropical Africa [8], [9]. The wood from the tree is used for indoor construction, light flooring furniture, canoes, carving, veneer and plywood. Various parts of *A. zygia* (bark, fruit, flowers and leaves) have been reported to be used as medicinal remedies [10]. The leaves and bark of this plant are used as traditional remedies in Africa for the treatments of fever, malaria, diarrhoea, deem, and conjunctivitis [11]. The powdered bark of *A. zygia* is used alone or as a decoction in southern Sudan as an antimalarial and antiparasitic drug [12]. In southern Nigeria, young *A. zygia* leaves are eaten as cooked vegetables, especially in soups [8]. Gum from the bark of the tree is used in the food industry as a stabilizer, in the cosmetic industry as a thickener and in the pharmaceutical industry as a drug coating agent [9].

Crateva adansonii DC. (*C. adansonii*) is a plant species of the family *Capparaceae* [13]. It is a small deciduous tree that can grow up to 10-15m tall. The plant can be found in many mainland African countries [14]. The bole is irregular and short, up to 50cm in diameter. The bark surface is smooth and grey to brown, while the inner bark is thin, yellow-brown with brown streaks [15], [14]

Various plant parts of *C. adansonii* are used in traditional medicine. In Cameroon, the plant is used to treat constipation, asthma, snake bites, post-menopausal complaints and cancer [16]. In Benin, the leaves and bark of this plant are used to treat jaundice, eczema, and rabies [17]. In Senegal, the roots are used in the treatment of sterility, syphilis, ear infections, yellow fever and jaundice [18]. *C. adansonii* leaves are used

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as vegetables in the Republic of Benin [19], [20], Burkina Faso [14] and Nigeria [21]. The present study was undertaken to determine the proximate, mineral and amino acid compositions of *A. zygia* and *C. adansonii* leaves and also to perform qualitative phytochemical screening on the methanol extract of the leaves. Findings from this study will add to existing information on the nutritional and medicinal values of these underutilized leafy vegetables.

II. MATERIAL AND METHODS

A. Collection of samples

Fresh plant leaves of A. zygia were collected from Ikere Ekiti, while the fresh leaves of *C. adansonii* were collected from Ado Ekiti, Ekiti State Nigeria. The leaves were identified in their fresh state by Mr. Felix Omotayo, the curator at the Herbarium of the Department of Plant Science, Ekiti State University, Ado-Ekiti.

B. Sample preparation

Plant leaves were air dried at room temperature for two weeks. The dried samples were pulverized by using a Marlex blender. The powdered leaves of each sample were separated into two, one part was kept in airtight containers prior chemical composition, mineral and amino acid analyses while the second portion was kept for phytochemical analysis.

C. Proximate analysis

The moisture, ash, fat, protein and crude fibre contents were determined using the methods of [22]

For ***the moisture content*** determination, clean and dry crucible was weighed, and the weight was recorded (W_1). Three grammes (3 g) of the sample was weighed into the crucible (W_2). The crucible with the sample was dried in the oven at 105 ^oC for three hours. The crucible was transferred to the desiccator to cool and the weight was noted. The process was continued until a constant weight (W_3) was obtained. The percentage loss in weight during drying was taken to be the percentage moisture content (Equation 1).

$$\% \text{ Moisture} = \frac{Weight\ loss}{Weight\ of\ sample} X100 \quad (1)$$

The ash content was determined by weighing 1 g of each sample into a clean, dried and previously weighed crucibles with lid (W_1). After removing the lid, sample was ignited over a low flame to char the organic matter. The crucible was then placed in a muffle furnace at 550 ^oC (lid removed). The ashing continued until a light grey or white ash was obtained. Crucible was then transferred directly into a desiccator, cooled and weighed immediately (W_2). The percentage ash content was obtained using Equation 2.

$$\text{Ash} = \frac{W_1 - W_2}{Weight\ of\ sample} \times 100 \quad (2)$$

The crude fat content was determined using Soxhlet extraction method. Two grammes (2 g) of sample was weighed into a filter paper. The filter paper with sample was folded neatly. Sample was thereafter placed inside a pre-dried thimble. Thimble with sample was inserted into the Soxhlet flask. A clean and dried boiling flask was weighed (W_1) and diethyl ether was poured into it. The boiling flask containing diethyl ether, Soxhlet flask with sample and condenser were assembled. Extraction was carried out under reflux for six hours. After extraction, the thimble was removed from the extraction barrel and dried. The solvent was distilled off and the boiling flask containing the fat was dried in the oven at a low temperature. The weight of the flask plus oil was recorded (W_2). Fat extracted from given quantity of sample was then calculated as the percentage fat content (Equation 3).

$$\%\text{Fat} = \frac{W_2 - W_1}{Sample\ Weight} \times 100 \quad (3)$$

The total nitrogen amount in the sample was determined following the micro Kjeldahl method. Digestion of the sample (2 g) was done in a Kjeldahl flask by boiling 20 mL of concentrated H_2SO_4 and a Kjeldahl digestion tablet until a clear mixture was obtained. The digest was filtered into 250 mL volumetric flask, made up to mark with distilled water and set up for distillation. Ammonia was steam-distilled from the digest to which 50 mL of 45% NaOH solution has been added. The distillate (150 mL) was collected into a conical flask containing 100 mL 0.1 N HCl and methyl orange was used as an indicator. The ammonia reacted with the acid in the receiving flask and percentage nitrogen (N) was estimated by back titration against 2 M NaOH. Nitrogen was calculated using the following equation.

$$\text{Nitrogen} = \frac{(A - B)X\ 1.4007}{Weight\ of\ sample} \times 100 \quad (4)$$

Where:

A = volume (mL) of standard HCl x normality of standard HCl

B = volume (mL) of standard NaOH x normality of standard NaOH

Percentage crude protein was obtained by multiplying the nitrogen value by a factor of 6.25%

Crude protein = Nitrogen in sample × 6.25.

To determine ***the crude fibre***, 2.5 g of each sample was extracted with diethyl ether in a Soxhlet apparatus. The extracted sample was air dried and transferred to a dry 1 L conical flask containing 1.25% sulphuric acid and was connected to a water cooled reflux condenser and boiled for exactly 30 minutes. The mixture was allowed to cool. It was then filtered through a clean white linen and washed with boiling water until the washings were no longer acid to litmus. The residue was further boiled with 1.25% sodium hydroxide solution. The flask was immediately connected to the reflux condenser and boiled for exactly 30 minutes. The mixture of the flask was filtered through the filtering cloth. Then the residue was thoroughly washed with boiling water and transferred to a Gooch crucible prepared with a thin compact layer of ignited asbestos. The residue was first thoroughly washed with hot water and then with about 15 mL of ethyl alcohol. The Gooch crucible and contents were dried at 105±2°C in an air oven until constant weight was achieved. The dried crucible and its contents were cooled and weighed. The contents of the Gooch crucible were incinerated in a

muffle furnace until all carbonaceous matter was burnt. The ash obtained was cooled in a desiccator and weighed.

The crude fibre was calculated as:

$$\text{Crude Fibre \% by weight} = \frac{W_1 - W_2}{W} \times 100$$

Where,

W_1 = weight in gramme of Gooch crucible and contents before ashing

W_2 = weight in gramme of Gooch crucible containing asbestos and ash

W = weight in gramme of the dried material taken for the test

Carbohydrate was determined by difference: {100 - (ash + moisture + crude protein + crude fibre + crude fat contents)}.

Gross energy value (kcal/100g) of the samples was obtained by multiplying crude protein content by 4, carbohydrate content by 4 and crude fat value by 9.

D. Phytochemical screening

For ***the phytochemical screening*** analysis, the powdered sample was transferred into a Soxhlet apparatus and was extracted in the Soxhlet extractor using methanol for 72 hours. The extract was concentrated to dryness and the residue obtained was stored and later used for phytochemical screening. Phytochemical screening for alkaloids, tannins, phenols, saponins, flavonoids, steroids, terpenoids and anthraquinones was performed by the method reported by [23], while cardiac glycosides and reducing sugar were determined by the method of [24].

E. Determination of mineral contents

Elemental analyses with the exception of sodium, potassium and phosphorus were carried out by Atomic Absorption Spectrometry (Bulk Scientific East Norwalk, CT, USA). Sodium and potassium were determined using a flame photometer (Corning, UK Model 405). KCl and NaCl were used to prepare the standards while phosphorus was determined by vanadomolybdate colorimetric method [25].

F. Sample preparation for amino acid analysis

About 2.0 g of sample was weighed into the extraction thimble and the fat was extracted with chloroform/methanol (2:1 v/v) mixture using a Soxhlet apparatus [22]. The extraction lasted for 5-6 h. About 30 mg of the defatted sample was weighed into glass ampoules. Seven millilitres of 6 M HCl was added and oxygen was expelled by passing nitrogen gas into the sample. The glass ampoules were sealed with a Bunsen flame and put into an oven at 105 ±5°C for 22 h. The ampoule was allowed to cool; the content was filtered to remove the humins. The filtrate was then evaporated to dryness at 40°C under vacuum in a rotary evaporator. The residue was dissolved with 5 ml acetate buffer (pH 2.0) and stored in a plastic specimen bottle and kept in the deep freezer.

G. Amino acid analysis

Amino acid analysis was by ion exchange chromatography (IEC) using the Technicon Sequential Multisample (TSM) Amino Acid Analyser (Technicon Instruments Corporation, New York). Detail of the procedure was given by [26]. The amino acid values reported were the averages of two determinations. Norleucine was the internal standard.

H. Determination of amino acid scores

Determination of the amino acid scores was first based on the formula given by [27]:

Amino acid score = amount of amino acid per test protein (mg/g) / amount of amino acid per protein in reference pattern (mg/g).

Secondly, the amino acid score was also determined based on the whole hen's egg profile [28]. Amino acid score was also calculated based on the composition of the amino acids obtained in the sample compared with the suggested pattern of requirements for pre-school children (2-5 years) [29].

I. Other determinations

The essential amino acid index (EAAI) was determined as described in literature [30]. The predicted protein efficiency ratio (P-PER) was determined using the equation

P-PER = –0.468 + 0.454 (Leu) – 0.105 (Tyr).

The Total amino acid (TAA), total essential amino acid (TEAA), total non-essential amino acid (TNEAA), total acidic amino acid (TAAA), total basic amino acid (TBAA), total neutral amino acid (TNAA) total sulphur amino acid (TSAA) and total aromatic amino acid (TArAA) and their percentage values, percentage cystine in TSAA (% Cys/TSAA), Leu/Ile ratios were calculated. ***The isoelectric point (pI)*** was calculated using the equation of the form [31]:

$$IP_m = \sum_{i=1}^{n} I_{P_i X_i}$$

Where IP_m is the isoelectric point of the mixture of amino acids, IP_i is the isoelectric point of the i^{th} amino acid in the mixture and X_i is the mass or mole fraction of the i^{th} amino acid in the mixture.

III. RESULTS AND DISCUSSION

A. Proximate composition

The proximate composition of the leaves investigated in this study, as presented in Table 1, revealed that the leaves of *A. zygia* contained higher values of moisture, crude protein, crude fat and crude fibre than *C. adansonii*. The respective moisture content recorded for both A. *zygia* and *C. adansonii* was 10.4 and 9.63 %. These values were lower than 14.71%, 13.7%, and 14.78% respectively reported for *Brillantaisia patul*a leaves [32] and *Adenia cissampeloides* leaves [33]. The relatively low moisture contents revealed that the studied plant leaves would be less prone to deterioration.

The crude protein values recorded for both plant leaves, 29.4% (*A. zygia*) and 18.3% (*C. adansonii*), compare favourably with 13.25 – 29.34% reported for lesser-known legumes in Nigeria [34]. However, these values were higher than 11.63% recorded for *Ficus cordata* leaves [35], 9.5 – 11.10% for *Cinnamon tamala* leaf varieties [36] and 14.36% for red bean [34]. Protein is required for essential body processes such as water balancing, nutrient transport and muscular contractions [37]. The crude protein contents of A. zygia and C. adansonii leaves indicate that they are good sources of protein.

The respective crude fat values of *A. zygia* and *C. adansonii* leaves were: 7.63 and 4.11%. These values were lower than 46.2% [38] and 23.33% [39] reported for calabash seed flour and *Acacia nilotica* seeds respectively. This implies that the leaves cannot be recommended for large scale

production of vegetable oil. The ash content of *A. zygia* leaves was 6.18% while the one obtained for *C. adansonii* was 13.2%. The ash content of any sample is an indicator of the mineral present in the sample. The vegetable leaves analyzed for in this study had average crude fibre contents of 5.84 (A. zygia) and 4.20% (C. *adansonii*). Fibre is significant in the prevention of diverticulosis, absorption of trace elements in the gut and in elimination of undigested food materials [40].

The carbohydrate values (40.6-50.6 %) obtained in this research were higher than 23.54 and 30.63 % reported for *Napoleona imperialis* and *Pterocarpus santalinoides*, two underutilized vegetables [41]. Carbohydrates are the primary source of energy for human beings and generally add to the bulk of the diet [42]. The calorific (energy) value of *A. zygia* leaves (349 kcal/100g) was higher than 313 kcal/100g obtained for *C. adansonii* leaves. Both samples had higher calorific values than those found in selected raw leafy vegetables having a range of 81.34 to 99.93 kcal/100g [43].

Table 1: Proximate composition of *A. zygia* and *C. adansonii* leaves and calculated energy values (kcal/100g)

Parameters	*A. zygia*	*C. adansonii*
Moisture	10.4±0.1	9.63±0.04
Crude Protein	29.4±0.0	18.3±0.1
Crude Fat	7.63±0.04	4.11±0.02
Ash	6.18±0.04	13.2±0.1
Crude Fibre	5.84±0.06	4.20±0.03
Carbohydrate	40.6±0.0	50.6 ±0.1
Energy	349	313

Values are reported as mean ± standard deviation of duplicate determinations

B. Phytochemical screening

Phytochemicals are non-nutrient bioactive components that are largely responsible for scavenging harmful radicals following oxidative stress by producing antioxidants [44]. Phytochemical screening serves as the initial step in predicting the types of potentially active compounds from plants [45]. Results of the phytochemical screening of methanol extract of *A. zygia* and *C. adansonii* leaves as shown in Table 2 revealed the presence of alkaloids, tannins, phenols, saponins and flavonoids in both samples. Terpenoids were present in *A. zygia* leaves but absent in C. adansonii leaves. Steroids and anthraquinones were absent in the two plant leaves while cardiac glycosides and reducing sugars were absent in *A. zygia* leaves but present in C. adansonii leaves. Phytochemicals are found in plants and their consumption generally provides beneficial health effects. Alkaloids have many pharmacological activities including antihypertensive and antiarrhythmic effects, antimalarial activity and anticancer actions [46]. Phenolic compounds are potent antioxidants and free radical scavengers which can act as hydrogen donors, reducing agents, metal chelators and singlet oxygen quenchers [47]. Saponins exhibit cytotoxic effects and growth inhibition against a variety of cells making them have anti-inflammatory and anticancer properties [48]. Flavonoids are effective antioxidants and free radical scavengers; they also protect the immune system [49].

Terpenoids show significant pharmacological activities, such as antiviral, antibacterial, antimalarial, anti-inflammatory, inhibition of cholesterol synthesis, and anti-cancer activities [50]. Cardiac glycoside is a typical secondary metabolite that stimulates the heart in case of heart failure [51]. The presence of these secondary metabolites in the *A. zygia* and *C. adansonii* leaves might have contributed to the rich medicinal value as well as the physiological activities of the plants.

Table 2: Phytochemical screening of methanol extract of *A. zygia and C. adansonii* leaves

Phytochemicals	*A. zygia*	*C. adansonii*
Alkaloids	+	+
Tannins	+	+
Phenols	++	++
Saponins	++	+
Flavonoids	+	+
Cardiac glycosides	-	+
Steroids	-	-
Anthraquinones	-	-
Reducing sugars	-	+
Terpenoids	+	-

+ indicates present ++ indicates moderately present - indicates absent

C. Mineral composition

The results of the macro and micro elemental compositions of the leaves of *A. zygia* and *C. adansonii* are presented in Table 3. Among the macrominerals studied, calcium had the highest concentration in both samples with *C. adansonii* having a higher value of 1210 mg/100g when compared to 930mg/100g obtained for *A. zygia* leaves. However, higher values were obtained in *A. zygia* leaves for potassium (841mg/100g), phosphorus (449mg/100g) and magnesium (165mg/100g) than in *C. adansonii* leaves with 589mg/100g, 222mg/100g, 44.7mg/100g for potassium, phosphorus and magnesium respectively. Moreover, sodium had the lowest concentration in both samples. Macrominerals play essential roles in the body, such as maintaining strong bones and teeth, synthesis of hormones and enzymes, muscle function, fluid balance etc. Calcium is vital in neuromuscular function, enzyme-mediated processes and blood clotting [52. Potassium is an important mineral for the proper functioning of all cells, tissues and organs of the body [53]. Phosphorus is an enzymatic component and is essential for proper immune function, energy metabolism and acid-base balance of the body [3]. Magnesium aids phosphorus absorption and its use also improve glucose tolerance [54]. Sodium regulates fluid balance in the body and helps in the proper functioning of muscles and nerves [55]. Iron was the most abundant micromineral in the two samples, but the concentration found in *C. adansonii* (51.7mg/100g) was more than 20.6mg/100g in *A. zygia* leaves. From the result, it could be deduced that *C. adansonii* leaves would be a better source of iron. Iron has been reported as an essential trace metal and plays numerous biochemical roles in the body, including oxygen binding in haemoglobin and acting as an important catalytic centre in many enzymes, for example, the cytochrome system [56]. Other trace minerals analyzed include manganese, zinc, and

copper. The concentration of zinc (3.81mg/100g) was the least in *A. zygia* leaves, while copper had the least concentration in *C. adansonii* leaves (0.81mg/100g). Manganese is involved in cellular reproduction, normal functioning of the immune system, maintenance of the blood sugar levels and digestion and bone growth [57]. Zinc plays a vital role in gene expression, regulation of cellular growth and participates as a co-factor in several enzymes responsible for carbohydrates, proteins and nucleic acids metabolism [58]. Deficiencies of copper can result in hernias, aneurysms and blood vessel breakage manifesting as bruising or nosebleeds [59]. Lead was not detected (N.D) in any of the two samples.

Calculated mineral ratios were also shown in Table 3. The Na/K ratio in the body is very important for the prevention of hypertension. To prevent high blood pressure, a Na /K ratio of 0.60 is recommended [60]. Values obtained for *A. zygia* and *C. adansonii* (0.033 and 0.063) leaves were less than 0.60 which is an indication that these plant leaves would not promote high blood pressure in the body. A food is considered good if the Ca/P ratio is above 1 and poor if the ratio is less than 0.5, while Ca/P ratio above 2 helps to increase the absorption of calcium in the intestine [61]. The ratio of Ca/P found in this study was within acceptable limits.

Table 3: Mineral composition of *A. zygia and C. adansonii* leaves

Minerals	***A. zygia* (mg/100g)**	***C. adansonii* (mg/100g)**
Sodium (Na)	27.9±0.0	36.9±0.1
Potassium (K)	841±1	589±2
Magnesium (Mg)	165±0	44.7±0.2
Phosphorus (P)	449±1	222±0
Iron (Fe)	20.6±0.2	51.7±0.0
Calcium (Ca)	930±2	1210±14
Zinc (Zn)	3.81±0.08	4.62±0.03
Manganese (Mn)	8.15±0.07	13.7±0.0
Copper (Cu)	4.21±0.04	0.81±0.01
Lead (Pb)	N.D	N.D
Na/K	0.033	0.063
Ca/P	2.07	5.45

Values are reported as mean ±standard deviation of duplicate determinations

D. Amino acid compositions of A. zygia and C. adansonii leaves

Table 4 presents the results of the amino acid concentration of *A. zygia* and *C. adansonii* leaves. Eighteen amino acids were present in *A. zygia* and *C. adansonii* leaves. Amino acids with high concentration (5.00g/100g cp and above) in *A. zygia* include: Glu, Asp, Lys, Leu, Pro, Ala, Gly, Phe, Val and Ile. The other eight amino acids present in *A. zygia* leaves had values below 5.00g/100cp. However, in *C. adansonii* leaves, only Glu, Asp, Leu and Lys had high concentrations of amino acids, the other 14 amino acids had values lower than 5.00g/100g cp. Amino acid with the highest concentration was Glu with *A. zygia* having 13.4g/100g cp and *C. adansonii* having 13.6g/100g cp. Coincidentally, both samples had the same level of Asp (10.4g/100g cp). Methionine was the amino acid with the lowest concentration in both leaves.

Table 5 shows the concentrations of TAA, TEAA, TNEAA, TAAA, TBAA, TNAA, TSAA, TArAA and their percentage values. Some other parameters such as Leu/Ile ratios, predicted protein efficiency ratio (P-PER), isoelectric point (pI) and essential amino acid index (EAAI) are also presented in Table 5. Although, the TAA obtained from the leaves of *A. zygia* (92.0g/100g cp) was higher than that of *C. adansonii*, reasonable amounts of amino acids were also present in *C. adansonii* leaves (82.7g/100g cp). The TAA present in the two plant leaves indicates that they would contribute significantly to the supply of amino acids in the diet.

The TEAA with His obtained in the present study for *A. zygia* and *C. adansonii* leaves were 42.3g/100g cp and 38.3g/100g cp, while the percent ratio of TEAA (with His) to TAA were 46.0 and 46.3 respectively. The percent ratio of TEAA (with His) to TAA of these samples compares favourably with the value recorded for the leaves of *Moringa oleifera* (46.4%) [62]; *Cucurbita maxima* (46.6%) [63] and cashew nut flour (43.7%) [64]. Essential amino acids are those that cannot be synthesized or those that are synthesized inadequately by the body relative to needs and hence must be obtained from diets to meet physiological requirements [65]. The results showed that the leaves are good sources of essential amino acids.

The concentration of TArAA present in *A. zygia* (9.37g/100g cp) and *C. adansonii* (7.36g/100g cp) leaves suggests that an appreciable amount of aromatic amino acids could be obtained from these vegetables since these values were within the range recommended for ideal infant protein (6.8-11.8g/100g cp) [29] TSAA of *C. adansonii* (2.49 g/100g cp) was slightly higher than that of *A. zygia* leaves (1.99 g/100g cp). The value for both samples was 42.9% (*C. adansonii*) and 34.3% (*A. zygia*) of 5.8g/100g cp recommended for infants [29]. The percent Cys in TSAA obtained for the leaves of *A. zygia* and *C. adansonii* compared favourably with values mostly prevalent in plant samples. The %TNAA in the samples was high and this indicates that it would form the bulk of the amino acid in the leaves. The protein present in *A. zygia* and *C. adansonii* was probably acidic in nature because the % TAAA was greater than % TBAA in both samples (Table 5). The Leu/Ile ratio values of the plant leaves in the present study (1.24 and 1.15) were better than the range of values of 0.832-1.15 reported for raw, steeped and germinated samples of *Zea mays* L. Dk 818 [66]. The P- PER value of the samples ranged between 2.01 (*A. zygia*) and 2.71 (*C. adansonii*). It has been reported that protein efficiency ratio (PER) value below 1.5 indicates a protein of poor quality; between 1.5 and 2.0 an intermediate quality and above 2.0 good quality proteins [67], [68]. From the results, *A. zygia* and *C. adansonii* leaves contain good quality proteins.

The pI of the leaves was 5.30 (*A. zygia*) and 4.70 (*C. adansonii*). The EAAI ranged from 1.09 in *C. adansonii* to 1.19 *A. zygia* leaves. EAAI is useful as a rapid tool to evaluate food formulations for protein quality, although it does not account for differences in protein quality due to various processing methods or certain chemical reactions [30].

Table 4: Amino acid concentrations of *A. zygia* and *C. adansonii* leaves (g/100g cp)

Amino acid	*A. zygia*	*C. adansonii*
Glycine (Gly)	5.35±0.12	4.54±0.14
Alanine (Ala)	5.72±0.19	4.67±0.1
Serine (Ser)	4.49±0.51	3.54±0.16
Proline (Pro)	5.83±0.23	4.38±0.0
Valine (Val)*	5.03±0.44	3.59±0.13
Threonine (Thr)*	4.45±0.14	3.20±0.07
Isoleucine (Ile)*	5.0 ±0.0	4.94±0.46
Leucine (Leu)*	6.21±0.04	7.36±0.91
Aspartic acid (Asp)	10.4±0.1	10.4±0.58
Lysine (Lys)*	9.12±0.39	6.51±0.18
Methionine (Met)*	0.798±0.007	0.82±0.01
Glutamic acid (Glu)	13.4±0.4	13.6±0.01
Phenylalanine (Phe)*	5.22±0.04	4.57±0.28
Histidine (His)*	2.27±0.06	1.53±0.05
Arginine (Arg)*	3.41±0.57	4.63±0.23
Tyrosine (Tyr)	3.35±0.00	1.62±0.07
Tryptophan (Trp)*	0.801±0.002	1.17±0.06
Cystine (Cys)	1.19±0.02	1.67±0.08
Total (TAA)	92.0	82.7

*Essential amino acid

Table 5: Summary of amino acid compositions of *A. zygia* and *C. adansonii* leaves

Parameters	*A. zygia*	*C. adansonii*
Total Amino Acid (TAA)	92.0	82.7
Percent total amino acid (%TAA)	100	100
Total non-essential amino acid (TNEAA)	49.7	44.4
Percent total non-essential amino acid (%TNEAA)	54.0	53.7
Total essential amino acid (TEAA) with Histidine	42.3	38.3
Percent total essential amino acid (%TEAA) with Histidine	46.0	46.3
Total essential amino acid (TEAA) without Histidine	40.0	36.8
Percent total essential amino acid (% TEAA) without Histidine	43.5	44.5
Total neutral amino acid (TNAA)	53.4	46.1
Percent total neutral amino acid (%TNAA)	58.0	55.7
Total acidic amino acid (TAAA)	23.8	24.0
Percent total acidic amino acid (% TAAA)	25.9	29.0
Total basic amino acid (TBAA)	14.8	12.7
Percent total basic amino acid (%TBAA)	16.1	15.4
Total sulphur amino acid (TSAA)	1.99	2.49
Percent total sulphur amino acid (%TSAA)	2.16	3.01
Percent cystine in TSAA	59.8	67.1
Total aromatic amino acid (TArAA)	9.37	7.36
Percent total aromatic amino acid (%TArAA)	10.2	8.90
Leu/Ile	1.24	1.49
Calculated isoelectric point (pI)	5.30	4.70
Predicted protein efficiency ratio (P-PER)	2.01	2.71
Essential amino acid index (EAAI)	1.19	1.09

Table 6: Essential amino acid scores of *A. zygia* and *C. adansonii* leaves based on [27] **standards**

Amino acid	[a]Suggested Level	Sample score (*A. zygia*)	Sample score (*C. adansonii*)
Ile	40	1.25	1.24
Leu	70	0.887	1.05
Lys	55	1.66	1.18
Met + Cys	35	0.569	0.711
Phe + Tyr	60	1.43	1.03
Thr	40	1.11	0.80
Trp	10	0.801	1.17
Val	50	1.01	0.718

[a][27]

The amino acid scores of *A. zygia* and *C. adansonii* leaves based on [27] standard is depicted in Table 6. In *A. zygia*, scores obtained for Lys, Phe + Tyr, Ile, Thr and Val showed that the amino acids were higher than the suggested values, while for Leu, Trp and Met + Cys the values were below the recommended values. The limiting amino acid in *A. zygia* leaves was Met + Cys with 0.569 score. However, in *C. adansonii* leaves, Ile, Lys, Trp, Leu and Phe + Tyr scores showed that the concentrations of these amino acids were

above the suggested values, while Thr, Val and Met + Cys had lower values than the recommended level and the least amino acid was Met + Cys and Val with 0.711 and 0.718 scores respectively. The scores obtained for the leaves of *A. Zygia* and *C. adansonii* based on this scoring pattern showed that both samples contained reasonable amounts of essential amino acids.

T**able 7**: Amino acid scores of *A. zygia* and *C. adansonii* leaves with respect to whole hen's egg profile[b]

Amino acid	Whole hen's egg (g/100g)	Sample score (*A. zygia*)	Sample score (*C. adansonii*)
Val	7.50	0.671	0.479
Thr	5.10	0.873	0.628
Ile	5.60	0.893	0.882
Leu	8.30	0.748	0.887
Lys	6.20	1.47	1.05
Met	3.20	0.249	0.256
Cys	1.80	0.661	0.928
Phe	5.10	1.02	0.896
Tyr	4.00	0.838	0.405
Trp	1.80	0.445	0.650
Gly	3.00	1.78	1.51
Ala	5.40	1.06	0.865
Ser	7.90	0.568	0.448
Pro	3.80	1.53	1.15
Asp	10.7	0.972	0.972
Glu	12.0	1.12	1.13
His	2.40	0.946	0.638
Arg	6.10	0.559	0.759

[b][28]

Table 7 reveals the amino scores of the leaves of *A. zygia* and *C. adansonii* based on the whole hen's egg values. From the results, Gly, Pro, Lys, Glu and Phe with scores greater than 1 in *A. zygia* had better values when compared with the whole hen's egg. Other amino acids in *A. zygia* had scores greater than 0.5 or 50% except for Met and Trp. Moreover, in *C. adansonii*, Gly, Pro, Glu and Lys had scores above 100% while Val, Ser, Try and Met had less than 50% scores. The other amino acids present in *C. adansonii* had scores above 50%. Scores based on the whole hen's egg profile revealed that Met was the least amino acid in *A. zygia* and *C. adansonii* with 0.249 and 0.256 scores respectively.

Table 8: Essential amino acid scores of *A. zygia* and *C. adansonii* leaves based on requirements of pre-school children (2-5 years) scoring pattern [29]

Amino acid	Pre-school (g/100g)	Sample score (*A. zygia*)	Sample score (*C. adansonii*)
Val	3.50	1.44	1.03
Thr	3.40	1.31	0.941
Ile	2.80	1.79	1.76
Leu	6.60	0.941	1.12
Lys	5.80	1.57	1.12
Met + Cys	2.50	0.796	0.996
Phe +Tyr	6.30	1.36	0.983
Trp	1.10	0.728	1.06
His	1.90	1.19	0.805

Table 8 shows the amino acid score of the samples based on the suggested amino acid requirement for the pre-school children. In *A. zygia* leaves, amino acid scores for Ile, Lys, Val, Phe + Tyr, Thr and His showed that the level of these amino acids exceeded the reference value for pre-school children, while Leu and Trp with lower scores were below the recommended values. Trp with the least score (0.728) was the limiting amino acid and would need a correction of 100/72.8 or 1.37. Amino acid scores for *C. adansonii* showed that Ile, Leu, Lys, Trp, Val were better than the recommended value for the preschool children, while Met + Cys, Phe + Tyr, Thr and His with lower scores had lower concentrations than the required values. His was the limiting amino acid with 0.805 score and would need a correction of 100/80.5 or 1.24.

IV. Conclusion

This study investigated the proximate, amino acid and mineral compositions and phytochemical screening of two indigenous African leafy vegetables; *A. zygia* and *C. adansonii*. The study revealed that the two leafy vegetables contained substantial amounts of crude protein and essential mineral elements needed for human and animal growth. The leaves contained eighteen amino acids. Glutamic and aspartic acids were the most abundant amino acids in the two plant leaves.

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